concentrations of other cobalamins in the media. Thus, the bioconjugates are still susceptible to decomposition when not bound to haptocorrin. While haptocorrin does stabilize the bioconjugates by several orders of magnitude, slow decomposition is still seen during the assay. However, this assay does indicate that the bioconjugates are stabilized such that a significant amount is still intact during the time of uptake and photolysis.

The data from the photolysis timecourse and activity assays are summarized in Figures 5-9 for compound **3** in each of the cell lines tested. Similar results were seen for compound **4**. In general, both bioconjugates showed similar uptake and photolysis behavior in the photolysis timecourse assay. The maximal photolytically induced toxicity is seen one hour after treatment with either of the bioconjugates. In all cases, photolysis of the bioconjugate demonstrated increased toxicity over that of unconjugated chlorambucil. Figure 5 shows that the chemotherapeutic drug, chlorambucil, has an LD_{50} of about 2 μM with respect to the HCT-116 cell line, whereas the bioconjugate shows no substantial toxicity at concentrations approaching 100 μM. If cells treated with the bioconjugate are subjected to brief irradiation with red light 12 hours after dosing, the LD_{50} decreases by a factor of 25 to 0.08 μM. If a 10-fold excess of vitamin B_{12} is added to saturate the cell surface receptors, the bioconjugate is not taken into the cells and photolysis triggers release of the active chlorambucil in the cell culture medium. The released chlorambucil now enters the cell by passive diffusion and an LD_{50} of 2 μM is observed — in close agreement with the value for chlorambucil standard.

Figure 6 shows that in cell line HL-60, the unconjugated chlorambucil standard exhibits an LD_{50} of 0.5 μM, but the bioconjugate is at least 2-fold better with an LD_{50} of 0.2 μM. The cytotoxicity of MC-121 against the leukemia cell line is still a dramatic result when compared with the absence of toxicity when the cellular uptake of the conjugate is out-competed by the addition of 10 equivalents of vitamin B_{12}. Similar results were obtained with Meth-A cells. The HL-60 and Meth-A cells have a high turnover rate, and in the case of Meth-A divide more rapidly than the other cell lines. These cells may, in fact, metabolize cobalamin at a faster rate than the other cell lines and thus release the chlorambucil in significant concentrations without photolysis. In order for this to be practical, however, cobalamin metabolism must occur before significant hydrolysis of chlorambucil moiety. It is reported that HL-60 cells are able to convert vitamin B_{12} into the other cobalamin forms efficiently (more quickly than normal lymphocytes) (Quadros and Jacobsen, 1995) and thus, would be able to efficiently release the conjugated

chlorambucil. In the other cell lines, however, the bioconjugates are essentially not toxic in non-photolytic conditions, which is a promising indication that these bioconjugates may not be toxic in normal somatic cells or healthy hematopoetic cells. IC_{50} values of the bioconjugates in both non-photolytic and photolytic conditions are summarized in Table 1.

TABLE 1

IC_{50} values (μM)

Cell Line	*HCT-116*	*HL-60*	*B-16*	*Meth-A*	*Rd-995*
Chlorambucil	20.8	5.8	1.4	1.8	1.1
Photolysis					
3	1.7		0.6	0.2	0.2
4	1.1		0.3	0.3	0.3
No Photolysis					
3		3.2		210.1	
4		8.9		84.2	

It will be appreciated that the methods and compositions of the instant invention can be incorporated in the form of a variety of embodiments, only a few of which are disclosed herein. It will be apparent to the artisan that other embodiments exist and do not depart from the spirit of the invention. Thus, the described embodiments are illustrative and should not be construed as restrictive.

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